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# Major morphological properties of the leaf of some Burley tobacco genotypes

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#### Summary

Due to a number of problems relating to the production of burley tobacco, the Tobacco Institute Zagreb has recently included creation of new cultivars of this tobacco type into its overall tobacco production programme. Some of its major morphological properties are being investigated. Thus, two-year investigations of certain important morphological properties of tobacco leaf have been carried out with four cultivars and five  $F_1$ -hybrids on the experimental field at Božjakovina.

The results obtained show that the genotypes under study were significantly different as regards the investigated morphological properties of the leaf. The highest increase in leaf number was achieved in the period from the end of June to the middle of July. All genotypes had a satisfactory number of leaves, except for the line cultivar Hy 71, as well as thickness and were, thus, classified as thin-bodied tobaccoes. As for leaf weight, the best results were obtained with the F<sub>1</sub>-hybrids Poseydon×BA1, Poseydon×TN 86 and Hy 71×BA1. A slightly flatter leaf angle was determined for the line cultivar Poseydon and the standard cultivar Čulinec. There was a high incidence of TMV infection in all cultivars in both years of investigation. The line cultivar Poseydon was highly susceptible to PVY.

Key-words: Burley tobacco, genotype, leaf morphological properties, TMV and PVY infection.

## Wesentliche morphologische Blatteigenschaften der Tabak-Genotypen des Typs Burley

### Zusammenfassung

Im Laufe der vergangenen Jahre arbeitete das Tabakinstitut Zagreb in Kroatien aufgrund einer Reihe von Problemen im Zusammenhang mit der Tabakproduktion des Typs Burley im Rahmen des Programms der Tabakproduktion auch an der Schaffung neuer Sorten dieses Tabaktyps. Im Rahmen dieses Programms werden unter anderem auch seine wesentlicheren morphologischen Eigenschaften untersucht. Zu diesen Zweck wurden auf dem Versuchsfeld in Božjakovina zweijährige Versuche bezüglich einiger wesentlicher morphologischer Eigenschaften des Tabakblatts mit vier Sorten und fünf  $F_1$ -Hybriden durchgeführt.

Die erzielten Ergebnisse zeigen, daß sich die untersuchten Genotypen im Hinblick auf die geprüften morphologischen Blatteigenschaften wesentlich unterscheiden. Der größte Zuwachs der Blattzahl wurde im Zeitraum von Ende Juni bis Mitte Juli erzielt. Alle Genotypen mit Ausnahme der Liniensorte Hy 71 hatten eine zufriedenstellende Blattzahl und Dicke und wurden deswegen dem dünnblättrigen Tabak zugeordnet. Im Hinblick auf das Gewicht des Blattes haben sich die F<sub>1</sub>-Hybride Poseydon×BA1, Poseydon×TN 86 und Hy 71×BA1 als die besten gezeigt. Einen etwas kleineren Winkel der Blattdeviation hatten die Liniensorte Poseydon und die Standardsorte Čulinec. In beiden Versuchsjahren war eine große Ansteckung aller Genotypen mit TMV zu erkennen. Die Liniensorte Poseydon war ausgesprochen empfindlich gegen PVY.

Schlüsselworte: Burley Tabak, Genotyp, morphologische Blatteigenschaften, Ansteckung mit TMV und PVY.

#### 1. Introduction

Tobacco (*Nicotiana tabacum* L.), a very plastic species, is grown all over the world. In the last hundred years, as long burley tobacco has been grown, a number of its properties have been investigated. Parallel to the definition of ecological requirements – climate and soil – and growth regionalization of this tobacco type, more attention is increasingly being paid to the variety and line research as a precondition of successful production and its wider spreading (WELACKY 1981, DEVERNA and AYCOCK 1983, POPOVIĆ et al. 1985, MILLER 1987, WILKINSON and RUFTY 1990). Despite indications of a crisis, production of burley tobacco is increasing worldwide, with the exception of the EU countries where a decrease of 0.5 % has been recorded (BAJTEK et al. 1993).

In recent years, burley production in Croatia has met less than 10 % of domestic demand. For this reason, the Tobacco Institute Zagreb has included a development programme of new burley cultivars for the growing conditions of Croatia into its overall tobacco production programme. Among others, certain morphological, economic and chemical properties are investigated (DEVČIĆ and BOLSUNOV 1975, DEVČIĆ et al. 1984, TRIPLAT 1984, TRIPLAT et al. 1994) and the germplasm prospective for the growing conditions in Croatia is selected.

As it is well known, tobacco is grown for its leaves. Therefore, the object of this work was to investigate some problems primarily related to the leaf as the main product of tobacco. The aim of testing the chosen genotypes was to:

1. determine some major morphological properties of the genotypes under study (number, thickness, weight and angle of leaves) as well as differences, if any, between them;

2. establish the rhythm of leaf appearance and growth during the growing season; and

3. determine tobacco infection by some major virus diseases.

# 2. Materials and methods

The trial, set up according to the randomized block method with four replications, involved nine Burley cultivars and was carried out on the experimental field of the Tobacco Institute Zagreb at Božjakovina in 1992 and 1993. Each trial plot had one tobacco row of 25 plants and was sized 10 m<sup>2</sup> (10 × 1 m). In both years of investigation, tobacco was planted on May 18. Standard agrotechnical measures for Burley tobacco were applied. No topping was performed. Burley genotypes were represented by four cultivars and some of their  $F_1$ -hybrids, as well as hybrids crossed with cultivar BA1. Thus, in addition to the standard cultivar Čulinec (Devčić and BOLSUNOV 1975), cultivars TN 86 from the U.S.A. (MILLER 1987), Hy 71 and Poseydon (Devčić et al. 1984) and  $F_1$ -hybrids Hy 71×TN 86, Hy 71×BA1, Poseydon×TN 86, Poseydon×BA1 and TN 86×BA1 were represented.

The investigated genotypes were partly different considering shape, base, top, surface, veins and colour of the leaf and development of suckers. It should be stressed that line cultivar Poseydon and the standard cultivar Čulinec produce no suckers in the growing season whereas other genotypes do, but only in the upper third of the plant, to a moderate to high extent.

During the investigation period, the rhythm of leaf appearance was monitored for each genotype on the whole trial at three week intervals. From the data obtained, the leaf increase between two measurements was calculated, as well as the average increase per days between the same measurements.

The insertion angle was measured on middle leaves of five successive, healthy and undamaged tobacco plants in a row in the central part of each plot in the full flowering phase.

After standard curing, the weight of five selected plants was determined by weighing and their thickness with a micrometre.

Tobacco mosaic virus (TMV) infection as well as potato virus Y (PVY) infection were monitored during the investigation period and the results are expressed in percent relative to the total number of plants.

The obtained data were statistically processed by the analysis of variance and by Duncan's test so as to check the significance of the differences between average values (Duncan 1955).

#### 3. Results and discussion

#### 3.1 Rhythm of leaf appearance

The leaf, the vegetative organ of the plant for which tobacco is grown, is actually a yield indicator. Selection is, therefore, aimed at creating such genotypes that will be satisfactory in terms of leaf formation and ripening, their number, and also leaf weight, thickness, surface area and shape, as well as the leaf insertion angle on the stalk. The time of leaf formation and development is of decisive importance for the economic and technological characteristics of tobacco. Optimal growing conditions should, therefore, be provided in the period when the economically most valuable middle leaves are formed and nutrients are translocated into upper leaf position. Under the growing conditions of this country this is the period from the end of June to most of July. In the last few years, attempts have been made to develop, through selection, earlier cultivars that would undisturbedly form leaves and ripen at the right time, which would facilitate curing and improve the quality.

Based on DUNCAN's test for the number of leaves appearing in different observation periods, statistically significant differences were determined between the genotypes tested (table 1).

At the time of the first observation in the first investigation year, there were no significant differences between the genotypes. The initial number of leaves was 5 to 6 for all genotypes. The second observation revealed significant differences between the genotypes under study. The  $F_1$ -hybrid Hy 71×TN 86 had the significantly highest number of leaves (16), while the significantly lowest

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Rhythm	of	leaf	appearance	e in	1992	and	1993

Genotype		Leaf number DMRT 5 % 1992				Increase in leaf number between two measurements			Average daily increase in leaf number between two measurements				
· ·	18.6.	9.7.	30.7.	20.8.	10.9.	I–II	II–III	III–IV	IVV	I–II	II–III	III–IV	IV-V
Čulinec	6 (a)	14 (c)	19 (de)	19 (d)	21 (de)	8	5	0	2	0.38	0.24	0.00	0.10
TN 86	5 (a)	14 (c)	20 (cd)	24 (a)	26 (abc)	9	6	4	2	0.43	0.28	0.19	0.10
Hy 71	5 (a)	12 (d)	16 (f)	19 (d)	20 (e)	7	4	3	1	0.33	0.19	0.14	0.05
Poseydon	5 (a)	14 (c)	20 (cd)	22 (bc)	26 (abc)	9	6	<b>2</b>	4	0.43	0.28	0.10	0.19
Hy 71×TN 86	6 (a)	16 (a)	22 (ab)	23 (ab)	25 (bcd)	10	6	1	<b>2</b>	0.47	0.28	0.05	0.10
Hy 71×BA1	6 (a)	15 (ab)	23 (a)	24 (a)	26 (abc)	9	8	1	2	0.43	0.38	0.05	0.10
Poseydon×BA1	6 (a)	14 (c)	21 (abc)	24 (a)	27 (ab)	8	7	3	3	0.38	0.33	0.14	0.14
Poseydon×TN 86	6 (a)	16 (a)	21 (abc)	23 (ab)	26 (abc)	10	5	2	3	0.47	0.24	0.10	0.14
TN 86×BA1	6 (a)	14 (c)	20 (cd)	23 (ab)	28 (a)	8	6	3	5	0.38	0.28	0.14	0.24
			1993										
	17.6.	8.7.	29.7.	19.8.	9.9.	I–II	II–III	III–IV	IV-V	I–II	II–III	III-IV	IV-V
Čulinec	5 (a)	13 (a)	21 (a)	23 (ab)	24 (c)	8	8	2	1	0.38	0.38	0.10	0.05
TN 86	6 (a)	12 (a)	19 (a)	23 (ab)	25 (b)	6	7	4	<b>2</b>	0.29	0.33	0.19	0.10
Hy 71	5 (a)	12 (a)	18 (a)	21 (c)	23 (d)	7	6	3	<b>2</b>	0.33	0.29	0.14	0.10
Poseydon	5 (a)	12 (a)	19 (a)	22 (bc)	25 (b)	7	7	3	3	0.33	0.33	0.14	0.14
Hy 71×TN 86	5 (a)	13 (a)	20 (a)	23 (ab)	24 (c)	8	7	3	1	0.38	0.33	0.14	0.05
Hy 71×BA1	6 (a)	14 (a)	21 (a)	23 (ab)	25 (b)	8	7	<b>2</b>	<b>2</b>	0.38	0.33	0.10	0.10
Poseydon×BA1	5 (a)	13 (a)	21 (a)	23 (ab)	25 (b)	8	8	2	2	0.38	0.38	0.10	0.10
Poseydon×TN 86	5 (a)	14 (a)	21 (a)	24 (a)	26 (a)	9	7	3	2	0.43	0.33	0.14	0.10
TN $86 \times BA1$	5 (a)	13 (a)	20 (a)	23 (ab)	25 (b)	8	7	3	$^{2}$	0.38	0.33	0.14	0.10

DMRT=DUNCAN's multiple range test

number (12) was recorded for the line cultivar Hy 71. At the time of the third and fourth observations, the  $F_1$ -hybrid Hy 71×BA1 had the significantly highest number of leaves, which means that this hybrid grows fastest. In the last observation, that is at the end of the growing season, most leaves (28) were recorded for the  $F_1$ -hybrid TN 86×BA1 and the fewest (20) for the line cultivar Hy 71, which had the fewest leaves throughout the whole growing season.

The results on the increase in leaf number between two measurements and the average increase per days indicate that the highest increase occurred between the 1st and 2nd, and between the 2nd and 3rd measurements. In the first case, an increase of 7 to 10 leaves was recorded. The highest average daily increase (0.47) was determined for  $F_1$ -hybrids Hy 71×TN 86 and Poseydon× TN 86. Between the 2nd and 3rd measurements, the highest increase of 8 leaves was that of the  $F_1$ -hybrid Hy 71×BA 1. The lowest average increase in most of the genotypes was recorded between the 4th and 5th measurements.

In the second investigation year, no significant differences between the genotypes were determined by the first three observations. The number of leaves recorded in the first observation ranged between 5 and 6, in the second observation 12 to 14, and in the third observation 18 to 21. The significantly highest numbers of leaves (24 and 26) at the time of the fourth and fifth observations, respectively, were recorded for the  $F_1$ -hybrid Poseydon×TN 86 while the lowest (21 and 23) were found for the line cultivar Hy 71.

The highest increase of nine leaves as well as the highest average increase (0.43) were achieved by the F<sub>1</sub>-hybrid Poseydon×TN 86 between the 1st and 2nd measurements. The increase between the remaining measurements varied from one to four leaves, depending on the genotype. The lowest average per day increase (0.05) was recorded between the 4th and 5th measurements for the F<sub>1</sub>-hybrid Hy 71×TN 86 and the standard cultivar Čulinec.

An overview of the results from both investigation years points to the conclusion that the genotypes differ significantly in the number of leaves and the rhythm of their appearance in the growing season. In both years, the highest increases were recorded for all genotypes in the period between the second half of June to the second half of July, that is in the optimal period for this cropping region. At the end of vegetation all genotypes had a satisfactory number of leaves, except for the line cultivar Hy 71.

Similar investigations, involving several cultivars and hybrids of yellow tobacco, were conducted in the Bosnian Sava Valley by BENKOVIĆ (1980). He reports on a faster leaf formation in hybrids so that the most valuable middle leaves are formed and developed at the optimal temperature, humidity as well as under favourable nutrient conditions, i.e. in June and July. According to DEVERNA and AYCOCK (1983) and WILKINSON and RUFTY (1990), F<sub>1</sub>-tobacco-hybrids grow faster and develop more leaves.

#### 3.2 Leaf weight

Leaf weight is also one of major parameters. The weight depends on the leaf thickness and on the population density. It is difficult to decide which of the two factors is more important (GARNER 1951). According to some investigations, the weight of leaf lamina increases with the leaf height on the plant and with tobacco ripeness (PRPIĆ 1977).

Leaf weight also depends on external factors, so it varied considerably due to changeable weather conditions in both investigation years. Significant differences in leaf weight between the tested genotypes were also recorded (table 2).

Table	e 2
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Genotype	Weight 1992	DMRT p=5 %	Genotype	Weight 1993	DMRT p=5 %
Poseydon×BA1 Poseydon×TN 86 Hy 71×TN 86 Hy 71×BA1 TN 86 TN 86×BA1 Poseydon Čulinec Hy 71	$\begin{array}{r} 4.80\\ 4.49\\ 4.16\\ 4.06\\ 3.99\\ 3.58\\ 3.40\\ 3.28\\ 2.37\end{array}$	a b cd cde f fg g h	Poseydon×TN 86 Poseydon×BA1 Hy 71×Ba1 Hy 71×TN 86 TN 86 Čulinec Poseydon TN 86×BA1 Hy 71	7.397.346.386.176.075.675.625.50 $4.47$	a ab c cd cde def defg efg h

Average leaf weight, g

DMRT=DUNCAN's multiple range test

In the first investigation year, the  $F_1$ -hybrid Poseydon×BA1 had the significantly highest leaf weight of 4.80 g. It was followed by the  $F_1$ -hybrid Poseydon×TN 86 with a leaf weight of 4.49 g. The differences between the F1-hybrids Hy 71×TN 86, Hy 71×BA1 and the cultivar TN 86 did not amount to statistical significance. The significantly lowest weight (2.37 g) was recorded for the line cultivar Hy 71, while the standard cultivar Čulinec was only negligibly better.

In the second investigation year, higher leaf weight values were obtained for all the genotypes under study, amounting to more than 2 g per leaf. The highest weight was recorded for the genotypes Poseydon×TN 86 and Poseydon×BA1 also in this year but, in contrast to the preceding year, without significant differences between them. They were followed by the same genotypes as in the first investigation year, viz. Hy 71×BA1, Hy 71×TN 86 and TN 86. Significantly lowest leaf weight was again recorded for the line cultivar Hy 71 (4.47 g) while the standard cultivar Čulinec achieved a higher, but still insufficient weight.

However, even the highest values for leaf weight should be considered as rather low. Unfortunately, the planned irrigation was not carried out for technical reasons, which would certainly have increased the total yield and the weight of each individual leaf in the drought conditions present in both years. According to the results obtained by CHANG and JOHNSON (1975), if tobacco is properly tended, which includes irrigation, it is possible to achieve a weight of about 8.5 g of lower leaves, up to 12 g of middle leaves, and 12 g of top leaves. It is not difficult to assume the benefits that would result from introduction of irrigation as a regular measure, primarily in extremely dry years.

#### 3.3 Leaf thickness

Leaf thickness is rather variable, mainly changing under the influence of site factors and agrotechniques, and, according to BURK et al. (1971), also the leaf position on the stalk. Based on their own research work in Macedonia, PEČIJA-RESKI and MIRČESKI (1978) maintain that changes of leaf thickness are negligible in comparison with leaf size. Our investigations were aimed at changes of leaf thickness in dependence on the genotype. Results of the average leaf thickness measurements are presented in table 3.

Note that the significantly thickest leaves in 1992 were obtained by the  $F_1$ -hybrids Poseydon×BA1 (0.0917 mm) and Poseydon×TN 86 (0.0915 mm). In most genotypes, the thickness ranged from 0.07 to 0.08 mm. The significantly

		5			
Genotype	Thickness 1992	DMRT p=5 %	Genotype	Thickness 1993	DMRT p=5 %
Poseydon×BA1 Poseydon×TN 86 TN 86 Hy 71×TN 86 Hy 71×BA1 TN 86×BA1 Čulinec Poseydon Hy 71	$\begin{array}{c} 0.0917\\ 0.0915\\ 0.0882\\ 0.0880\\ 0.0841\\ 0.0815\\ 0.0812\\ 0.0791\\ 0.0634 \end{array}$	a b cd f fg h h	Poseydon×BA1 Poseydon×TN 86 Hy 71×BA1 Hy 71×TN 86 TN 86 Čulinec TN 86×BA1 Poseydon Hy 71	$\begin{array}{c} 0.0786\\ 0.0915\\ 0.0767\\ 0.0767\\ 0.0747\\ 0.0745\\ 0.0744\\ 0.0724\\ 0.0594 \end{array}$	a b cd de def defg h h

Table 3 Average leaf thickness.mm

DMRT = DUNCAN's multiple range test

thinnest leaves were those of the line cultivars Poseydon and Hy 71. The standard cultivar Čulinec was significantly better only than the last two cultivars.

Taken as a whole, all tested genotypes had thinner leaves in 1993 than in 1992. Also here, the  $F_1$ -hybrids Poseydon×BA1 and Poseydon×TN 86 had significantly thickest leaves. In the relative sense, there are small differences in leaf thickness between the genotypes under study in relation to the previous year. In most genotypes, the thickness ranged between 0.072 and 0.076 mm. Line cultivars Poseydon and Hy 71 had significantly thinnest leaves also in 1993.

A comparison of our results with those of some earlier investigations (Aran-DELOVIĆ et al. 1973, POPOVIĆ et al. 1985) allows the conclusion that the leaf thickness of this tobacco type varies in the range from 0.06 to 0.11 mm. According to the existing criteria for leaf thickness, on the basis of the results obtained, they are classified as thin-leaved tobaccoes.

# 3.4 Leaf angle

The leaf angle to the stalk defines the plant habit and it is determined by the tobacco types and genotypes (GARNER 1951). For the middle leaf of Virginia and Burley tobacco, this angle is between 40 and 60°.

Based on Duncan's test, statistically significant differences were determined between the genotypes tested in both investigation years (table 4).

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Genotype	Angle 1992	DMRT p=5 %	Genotype	Angle 1993	DMRT p=5 %
TN 86×BA1 Poseydon×TN 86 TN 86 Hy 71 Hy 71×TN 86 Hy 71×BA1 Poseydon×BA1 Čulinec Poseydon	58 57 57 57 55 48 47 45	a ab ab ab abc d d d	TN 86 Hy 71 Hy 71×TN 86 TN 86×BA1 Hy 71×BA1 Poseydon×TN 86 Poseydon×BA1 Čulinec Poseydon	57 57 56 55 55 50 48 47	a ab ab abc abc d d d

Table 4 Average leaf angle

DMRT=DUNCAN's multiple range test

In 1992, the largest leaf angle of 58° was recorded for  $F_1$ -hybrids TN 86×BA1 and Poseydon×TN 86. There were no statistically significant differences between these and the genotypes TN 86, Hy 71 and Hy 71×TN 86. The smallest angle of 45° was that of the line cultivar Poseydon, between which and the  $F_1$ -hybrid Poseydon×BA1 and the standard cultivar Culinec there were no statistically significant differences.

In 1993, genotypes TN 86 and Hy 71 had the largest leaf angle of 57°. F<sub>1</sub>-hybrids Hy 71×TN 86, TN 86×BA1, Hy 71×BA1 and Poseydon×TN 86 were not significantly different from the first two genotypes. It was also in this year that the smallest leaf angles were recorded for the same genotypes, viz. Poseydon×BA1, Culinec and Poseydon.

A comparison of the results from both investigation years shows that there are no large differences in the leaf angles in the same genotypes.

Similar investigations were conducted by DEVČIĆ and BOLSUNOV (1975) on the standard cultivar Čulinec. The leaf angle of the middle leaf was 48°, which is in agreement with our results.

# 3.5 Tobacco Mosaic Virus (TMV) and Potato Virus Y (PVY) infection

In recent years, virus diseases have caused serious damage to tobacco, primarily with respect to quality. The potato virus Y is certainly most dangerous. Since resistance to PVY is controlled by a recessive gene (Lucas 1975), parents possessing satisfactory resistance should be selected in order to obtain resistant  $F_1$ -hybrids (TRIPLAT et al. 1994).

The percent of TMV and PVY infected plants recorded in our investigations is given in table 5.

	TMN	D1717	<b>ETIN #X 7</b>	D1717
Genotype	1992	1992	1993	PVY 1993
Čulinec TN 86 Hy 71 Poseydon Hy 71×TN 86 Hy 71×BA1 Poseydon×BA1 Poseydon×TN 86	17.97 13.68 12.08 10.98 12.35 12.50 30.58 28.08	5.61 2.10 0.00 21.95 0.00 6.81 3.52 5.61	$14.44 \\ 12.94 \\ 20.00 \\ 32.94 \\ 14.81 \\ 10.45 \\ 22.89 \\ 27.58$	$\begin{array}{c} 3.33\\ 0.00\\ 0.00\\ 14.11\\ 0.00\\ 0.00\\ 3.61\\ 1.14 \end{array}$

Table 5Percent of TMV and PVY infected tobacco plants

In 1992, all the genotypes tested were TMV infected. The highest infection incidence was determined in the  $F_1$ -hybrids Poseydon×BA1 (30.58%) and Poseydon×TN 86 (28.08%). In most of the other genotypes, TMV infection ranged between 10 and 13%. In contrast to the line cultivar Poseydon, which is very susceptible to PVY, the genotypes Hy 71, Hy 71×TN 86 and TN 86×BA1 were not infected. Neither in other genotypes there was a large extent of infection.

Also in 1993, all the genotypes tested were TMV infected. The highest percent of infected plants was recorded in genotypes Poseydon (32.44%), Poseydon×TN 86 (27.58%) and Poseydon×BA1 (22.89%). In most of the other genotypes, TMV infection ranged between 10 and 15%. Analogously to the year

before, there were no signs of PVY infection in the genotypes Hy 71, Hy  $71 \times TN$ 86 and TN 86×BA1, and TN 86 and Hy 71×BA1.

High incidence of virus infection, particularly by TMV, was recorded in both investigation years. However, the disease mainly attacked top leaves, so it was not strongly reflected in the economic and technological characteristics of tobacco. The line cultivar Poseydon was markedly susceptible to PVY whereas the TN 86 cultivar, which is resistant to some PVY strains (REDDICK et al. 1991). manifested satisfactory resistance. The standard cultivar Culinec is very susceptible to TMV and slightly less to PVY.

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